PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 59.82765/001	**FOR FURTHER ACTION** See Form PCT/IPEA/416	
International application No. PCT/GB2004/005365	International filing date *(day/month/year)* 20.12.2004	Priority date *(day/month/year)* 19.12.2003

International Patent Classification (IPC) or national classification and IPC
C12N5/00, C12N5/10, C12N5/06, C12N5/08, A61K35/12, A61K48/00, G01N33/00

Applicant
OMNICYTE LTD et al.

1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.
2. This REPORT consists of a total of 7 sheets, including this cover sheet.
3. This report is also accompanied by ANNEXES, comprising:
   a. ☐ *sent to the applicant and to the International Bureau)* a total of sheets, as follows:
      ☐ sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).
      ☐ sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.
   b. ☐ *(sent to the International Bureau only)* a total of (indicate type and number of electronic carrier(s)) , containing a sequence listing and/or tables related thereto, in computer readable form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).

4. This report contains indications relating to the following items:

☒	Box No. I	Basis of the opinion
☒	Box No. II	Priority
☐	Box No. III	Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
☐	Box No. IV	Lack of unity of invention
☒	Box No. V	Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
☐	Box No. VI	Certain documents cited
☐	Box No. VII	Certain defects in the international application
☒	Box No. VIII	Certain observations on the international application

Date of submission of the demand	Date of completion of this report
11.07.2005	20.03.2006

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INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.
PCT/GB2004/005365

Box No. I Basis of the report

1. With regard to the **language**, this report is based on the international application in the language in which it was filed, unless otherwise indicated under this item.

   ☐ This report is based on translations from the original language into the following language , which is the language of a translation furnished for the purposes of:
   - ☐ international search (under Rules 12.3 and 23.1(b))
   - ☐ publication of the international application (under Rule 12.4)
   - ☐ international preliminary examination (under Rules 55.2 and/or 55.3)

2. With regard to the **elements*** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report)*:

Description, Pages

1-38	as originally filed

Claims, Numbers

1-41	as originally filed

Drawings, Sheets

1/16-16/16	as originally filed

☐ a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing

3. ☐ The amendments have resulted in the cancellation of:
   - ☐ the description, pages
   - ☐ the claims, Nos.
   - ☐ the drawings, sheets/figs
   - ☐ the sequence listing *(specify)*:
   - ☐ any table(s) related to sequence listing *(specify)*:

4. ☐ This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).
   - ☐ the description, pages
   - ☐ the claims, Nos.
   - ☐ the drawings, sheets/figs
   - ☐ the sequence listing *(specify)*:
   - ☐ any table(s) related to sequence listing *(specify)*:

* *If item 4 applies, some or all of these sheets may be marked "superseded."*

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.
PCT/GB2004/005365

Box No. II Priority

1. ☐ This report has been established as if no priority had been claimed due to the failure to furnish within the prescribed time limit the requested:
   ☐ copy of the earlier application whose priority has been claimed (Rule 66.7(a)).
   ☐ translation of the earlier application whose priority has been claimed (Rule 66.7(b)).

2. ☐ This report has been established as if no priority had been claimed due to the fact that the priority claim has been found invalid (Rule 64.1). Thus for the purposes of this report, the international filing date indicated above is considered to be the relevant date.

3. Additional observations, if necessary:

   see separate sheet

Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	1-41
	No:	Claims	none
Inventive step (IS)	Yes:	Claims	1-41
	No:	Claims	none
Industrial applicability (IA)	Yes:	Claims	1-18, 23-32, 37-41
	No:	Claims	no opinion: 19-22, 33-36

2. Citations and explanations (Rule 70.7):

   see separate sheet

Box No. VIII Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

see separate sheet

Re Item II

Priority

Claim 11 of the present application refers to deposited cells. In view of the description these cells were only deposited between the priority date and the filing date (see page 8, l.14 to 18). Thus, any subject-matter based on these cells or on a reference thereto does not enjoy priority rights.

Re Item V

Reasoned statement with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

Reference is made to the following document/s/:

D1: WO 01/71016 A (STEMCELLS, INC; LAGASSE, ERIC; WEISSMAN, IRVING, L) 27 September 2001 (2001-09-27)

D2: BUCALA R ET AL: "CIRCULATING FIBROCYTES DEFINE A NEW LEUKOCYTE SUBPOPULATION THAT MEDIATES TISSUE REPAIR" MOLECULAR MEDICINE, BLACKWELL SCIENCE, CAMBRIDGE, MA, US, vol. 1, no. 1, November 1994 (1994-11), pages 71-81, XP002051352 ISSN: 1076-1551

D3: VERFAILLIE C M: "Adult stem cells: Assessing the case for pluripotency" TRENDS IN CELL BIOLOGY, ELSEVIER SCIENCE LTD, XX, vol. 12, no. 11, November 2002 (2002-11), pages 502-508, XP002268623 ISSN: 0962-8924

D4: WO 02/064748 A (FURCHT, LEO, T; VERFAILLIE, CATHERINE, M; REYES, MORAYMA) 22 August 2002 (2002-08-22)

D5: DEANS ROBERT J ET AL: "Mesenchymal stem cells: Biology and potential clinical uses" EXPERIMENTAL HEMATOLOGY, NEW YORK, NY, US, vol. 28, no. 8, August 2000 (2000-08), pages 875-884, XP002201188 ISSN: 0301-472X

D6: JIANG Y ET AL: "PLURIPOTECNY OF MESENCHYMAL STEM CELLS DERUVED FROM ADULT MARROW" NATURE, MACMILLAN JOURNALS LTD. LONDON, GB, vol. 418, no. 6893, 4 July 2002 (2002-07-04), pages 41-49, XP001204372 ISSN: 0028-0836

D7: JAVAZON ELISABETH H ET AL: "Mesenchymal stem cells: Paradoxes of passaging" EXPERIMENTAL HEMATOLOGY (NEW YORK), vol. 32, no. 5, May 2004 (2004-05), pages 414-425, XP002321064 ISSN: 0301-472X, cited as P-document.

1. Novelty

The subject-matter of claims 1 to 41 is considered new in the sense of Article 33(2) PCT.

1.1 Claim 1 is directed to CD34 positive stem cells which are further defined by several functional features, such as self regeneration, capability of differentiation into ectodermal, mesodermal and endodermal cells and by their ability to adhere to tissue culture plastic. Dependent claims 2 to 16 define this stem cells by further functional or technical features of the stem cells or their progeny. Claim 17 is directed to a stem cell population defined by the process it is obtained by. Claim 18 is directed to a cell culture comprising such a stem cell population and a medium supporting its growth.

1.2 Example 1 of D1 shows that murine c-kit$^+$ Thy-1.1^{lo} Lin$^{-/lo}$ Sca-1$^+$ haematopoietic stem cells (HSC) transplanted into irradiated mice give rise to the haematopoietic system and to hepatocytes. Human CD34$^+$ Thy-1$^+$ Lin$^-$ HSCs are considered equivalent to said murine HSCs, however no such cells were tested for their developmental potential neither in vitro nor in vivo.

1.3 Apparently only 1% of CD34$^+$ cells have the ability to adhere to tissue culture plastic (see application, p.23, l.5-8). Thus, even if human HSCs had the same potential to develop to haematopoietic cells and hepatocytes they have to be considered as a non homogenous population of cells which may also comprise some cells having the characteristics of the stem cells of the present application.

1.4 D2 discloses CD34 and CD45 positive, CD38 negative, CD3 negative, CD19 negative, collagen positive and vimentin positive stem cells derived from peripheral blood which adhere to tissue culture plastic and proliferate in culture. However, the adherent cells represent a mixed population of CD34+ fibrocytic cells and non-proliferating monocytes which decline after 2 to 4 weeks of culture. Thus, D2 is considered to provide a non-homogenous cell population. The CD34 positive cells therein appear to develop a fibrocyte-like morphology both in culture and in implanted wound chambers. These fibrocytic cells are considered to be pluripotent precursors of fibroblast, smooth muscle or osteogenic lines at wound sites (for references see search report). Differentiation into endodermal or ectodermal cells was not studied.

1.5 Thus, the isolated stem cell population and cell culture according to claims 1 to 18 may be considered novel over the prior art, in particular in view of D1 and D2. Same applies to methods for their isolation according to claims 19 to 22, to methods using said stem cells (see claims 23 to 25, 33, 37 to 41) and to cell populations according to claims 26

to 32 and 34 to 36.

2. Inventive step

The subject-matter of claims 1 to 41 is considered inventive step in the sense of Article 33(3) PCT.

2.1 The application provides pluripotent stem cells derived from haematopoietic tissue (see claims 1 -18) as well as a method of isolating these (19-22).
Document D5, which is considered to represent the most relevant state of the art, discloses the isolation of mesenchymal stem cells (MCS) from haematopoietic tissue. The method of isolation disclosed by D5 differs from the method used in the application (see claims 20 to 22) by lacking steps (ii) (iii) consisting of the selection of CD34 positive cells after density gradient separation. The stem cells disclosed by D5 are MSC, whose developmental potential appears to be restricted to the mesenchymal lineage.

2.2 The problem to be solved by the present invention may be regarded as isolating pluripotent stem cells from haematopoietic tissue which have the potential to differentiate not only into mesodermal cells but also into endodermal and ectodermal cells.
The solution provided by the application is an additional selection of CD34 positive cells between density gradient separation and plating on a solid support. The resulting cell population ASC 34 was characterized in respect to tissue specific markers and morphology of cultured progeny. It appears that ASC 34 cells have indeed the potential to differentiate into cells of all three lineages.

2.3 A number of methods to isolate haematopoietic progenitor cells or mesenchymal adult stem cells are known in the art (see e.g. D3 to D7). As CD34 is generally considered as a marker for progenitors of the haematopoietic system or of other mesenchymal cells which are already determined to a certain lineage it is not considered obvious to introduce such a step in order to isolate pluripotent cells.

2.4 The solution to this problem proposed in claims 1 to 22 of the present application is therefore considered as involving an inventive step (Article 33(3) PCT).

2.5 Same applies to methods using said stem cells (see claims 23 to 25, 33, 37-41) and to cell populations according to claims 26 to 32 and 34 to 36.

3. Industrial applicability

Claims 19 to 22 and 33 to 36 are directed to methods of medical treatment or to a method comprising a surgical step (e.g. taking a blood sample). For the assessment of the present

said claims on the question whether they are industrially applicable, no unified criteria exist in the PCT Contracting States. The patentability can also be dependent upon the formulation of the claims. The EPO, for example, does not recognize as industrially applicable the subject-matter of claims to the use of a compound in medical treatment, but may allow, however, claims to a known compound for first use in medical treatment and the use of such a compound for the manufacture of a medicament for a new medical treatment.

Re Item VIII

Certain observations on the international application

The application does not meet the requirements of Article 6 PCT, because claims 1 to 16, 18, 19, 24 to 41 are not clear.

Claims 1, 19, 23 and claims 2 to 16, 18, 19, 24 to 41 referring thereto do not meet the requirements of Article 6 PCT in that the matter for which protection is sought is not clearly defined. The claims attempt to define the subject-matter in terms of functional features which rather describe the result to be achieved, i.e. the developmental or regenerative potential of the cells, which merely amounts to a statement of the underlying problem, without providing the technical features necessary for achieving this result, e.g. the steps necessary in order to obtain these cells.

It was considered that claim 26 was meant to be directed to a cell population produced by a method according to claims 19 to 22 and 25. The products of methods according to claims 23 to 24 could obviously not be considered novel over any known differentiated cell.